Is the consumption of Tilapia zillii and Clarias gariepinus from Edku Lake – El Beheira Governorate – Egypt safe for humans: a physiological and biochemical study and determination of five trace elements

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Research Article

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Abstract

The Egyptian government seeks to develop the northern lakes, including Edku Lake in the El Beheira governorate, to enhance the physiological functions of fish, and thus, improve their quality and promote their products. Therefore, this study was performed on the two cheapest fish species *T. zillii* and *C. gariepinus* in the Egyptian market, by studying proximate body composition, antioxidant enzyme activity, and bioaccumulation of some trace elements. In this study, *T. zillii* showed the highest mean of carbohydrates (16.11± 0.67 mg/g), fats (24.21 ± 0.93mg/g), ash (7.33 ±1.00 %), and water content (78.33 ± 1.85 %) whereas, the highest mean of protein content was recorded in *C. gariepinus* (64.33 ±1.01 mg/g). Antioxidant activity GSH (41.66 ± 2.01mg/g), CAT (21.66 ± 2.6 U/g), GPx (36.5 ± 3.01 mU/L), and GR (41.16 ± 1.0 U/L) in the liver of *T. zillii* demonstrated insignificant decrease, except GSH and CAT was significant at ($p < 0.05$). SOD activity showed an insignificant decrease in *T. zillii* (47±3.5 U/g). Concerning trace element concentrations, *C. gariepinus* demonstrated an increase in five trace elements than *T. zillii.* The abundance of the elements is in the order as Hg< Al < Cd <Pb < As in *T. zillii* and *C. gariepinus* muscles. The highest As was recorded in *C. gariepinus* (1.54 ± 0.08 µg/g), whereas, the lowest Hg was recorded in *T.zillii* (0.47 ± 0.08 µg/g). In this research, the concentrations of the trace element in the two studied fish species were above the limits permitted by EOSQC, FAO/WHO, and the EU'. The estimated weekly intake (EWI) of the elements concentrated in the two fish species was unsafe for young people (40 Kg), and children (15 Kg), whereas it was safe for consumption by adults (70 kg) with advice on safe levels of consumption for *C. gariepinus* for adults. So, this study recommended that the children should consume less than 16.12857 g/day or 112.90 g/week *T.zillii* muscle and 13.27 g/day or 92.92 g/week C*. gariepinus* muscle. Moreover, Youth should consume less than 43.01 g/day or 301.7g/ week *T.zillii* muscle and 35.39714 g/day or 247.78 g/week C*. gariepinus* muscle. Finally, adult should consume less than 23.1 g/day and 161.7 g/week *T.zillii* muscle, and 61.94286 g/day or 433.6 g/week *C. gariepinus* muscle to maintain their safety.

Introduction

Unnatural quantities of trace elements such as Cd, Pb, Hg, As and Al have been released to the aquatic environment via storm water and wastewater discharges following industrialization (Qin et al. 2015). They are a global concern due to their potential hazard effect and concentrated capacity in the aquatic environment. Most coastal lagoons are in danger, suffering from excessive commercial, agricultural, and domestic wastewater stream discharge (UNEP 1999; Batvari et al. 2013).

Edku Lake has the smallest area compared to Burullus and Manzalah Lakes, linked to the Mediterranean Sea via Bughaz El Meadia lake-sea connection (Fig. 1) (Shakweer 2006). According to EEAA (2017), the lake received vast amounts of drainage reached to 1.738 × 1012 m3/year, the primary source of toxic elements in aquatic ecosystems. (Renieri et al. 2014; EEAA 2017).

Fish has been considered a significant component of a balanced, healthy diet containing high-quality proteins, vitamins, and several other essential nutrients (Pieniak et al., 2010). Moreover, fish is a major source of polyunsaturated omega 3 fatty acids (PUFAs) whose benefits reduce inflammatory diseases, coronary heart disease, cancer, arterial hypertension, and contributes to normal neurodevelopment in children (Mozaffarian and Wu, 2011; Swanson et al. 2012). Furthermore, The American Heart Association has recommended fish consumption at least twice a week to achieve daily consumption of omega-3 fatty acids (Kris-Etherton et al. 2002).

Tilapia and African catfish are commercially valuable fish because, in several African countries, including Egypt, they are considered a cheap food source (Olmedo et al. 2013). Catfish and tilapia species contribute to over 80% of Lake Edku production (GAFRD 2012). These fish species seem to have been acclimatized to the prevailing conditions and to be able to survive, forming the main fish population in the lake.

In living organisms, the trace elements such as (Cd, Pb, Hg, As and Al) have no known significant role; cause extreme toxic effects even at deficient concentrations to all living organisms in particular human health (Sarmiento et al. 2011). Toxic effects occur when the mechanisms of metabolic, storage, detoxification, and excretory can no longer help counter uptake (Obasohan et al. 2008). Eventually, lead to physiological and biochemical changes (Olabanji and Oluyemi 2014; Abdel-Kader and Mourad 2020).

Several researchers in the literature have shown the toxicity of trace elements such as (Cd, Pb, Hg, Al, and As) on various fish species in the freshwater lakes such as *Oreochromis niloticus* fish from Lake Burullus, Egypt (Olabanji and Oluyemi 2014); freshwater fish *Labeo rohita* from Yellamallappa Chetty Karnataka (Noor and Zutshi 2016); *O. niloticus* collected from different locations from Egypt such as Mansoura and Abassa (El-Sappah et al. 2012); *C. gariepinus* fish from two basins at Lake Maryout, Egypt (Abdel-Kader and Mourad 2019a), tilapia species and *C. gariepinus* from Burullus Lake-Egypt (Abdel-Kader and Mourad 2020).

Proximate body composition is the analysis of water, fat, protein, and ash contents of fish (Love 1970). The biochemical profile in fish is a sensitive index of fish metabolism assessment under metallic stress. Several studies demonstrated that fish exposed to toxic elements such as Cd, Pb, Hg, As and Al recorded low levels of proteins and lipids (Sobha et al. 2007; Selvam et al. (2014); Ayanda et al. 2018; Abdel-Kader and Mourad 2019b; Abdel-Kader and Mourad 2020).

Toxic elements such as Cd, Pb, Hg, Al, and As reduce the activity of antioxidants, particularly which having the thiol group (—SH). Toxic elements generate reactive oxygen species (ROS) such as superoxide radical (O_2^-), hydroxyl radical (HO^-), and hydrogen peroxide (H_2O_2) (Romeo et al., 2000; Atli and Canli, 2010). Increased ROS production can destroy the defenses of the antioxidant of cells and result in a condition is known as “oxidative stress” and, eventually, death of cells (Ercal et al. 2001). The antioxidant defense systems of fish tissues, particularly the liver, protect them against oxidative stress (Basha and Rani, 2003; Atli and Canli, 2007). Several researchers estimated the activity of the antioxidant enzymes after exposure to toxic elements such as Cd, Pb, Hg, Al, and As in the different fish species collected from different freshwater lakes (Barata et al. 2005; Monteiro et al. 2010; Abdelazim et al. 2018; Elarabany and Bahnasawy 2019; Abdel-Kader and Mourad 2019b; Abdel-Kader and Mourad 2020), they recorded a reduction in the activity of the antioxidant enzymes after Cd, Pb, Hg, As and Al exposure.

The purpose of this study was (1) to determine the concentrations of five trace elements Cd, Pb, Hg, As and Al accumulated in the muscles of *T.zillii* and *C.gariepinus* collected alive by the fishermen from Edku Lake, Beheira Governorate, Egypt, these concentrations were compared with available certified safety guidelines published by the World Health Organization (WHO) and the Food and Agricultural Organization (FAO), European Community (EC), (2) to calculate the human risk assessment of intake of heavy metal at human health through consumption of the *T.zillii* and *C.gariepinus* muscles, (3) to check the proximate body composition in tissues samples *T.zillii* and *C.gariepinus* and, (4) finally, to determine the activity of the antioxidant enzyme of the liver of *T.zillii* and *C.gariepinus*.

Materials And Methods

Edku Lake is located 30 km northeast of Alexandria, one of the four coastal delta lakes linked to the Mediterranean Sea (Fig. 1). 30º 30 $\text{and } 30º23$ E and 31º 10 $\text{and } 31º18$ N. the area of the lake is 126 km^2, a depth is 50-150 cm. via primary drains, El-Khairy plus Barseek, it receives huge quantities of drainage water. El-Khairy Drain's water sources come from three drains known as El-Bousely, Edku and Damanhour sub drains, residential, agricultural as well as industrial waste transportation plus irrigation water from over 300 fish farms. Barseek drain carries farm drainage water to the lake (Badr and Hussein 2010). The area has diminished from 28.5x 10^3 to approximately 12x10^3 Fadden (Okbah and El-Gohary 2002). Due to land reclamation, large areas of approximately 150 km^2 have vanished (Badr and Hussein 2010).

Fish sample collection

The medium-sized fish from different sites from the Edku Lake — Egypt of *Tilapia zillii* and catfish *Clarias gariepinus* (six from each) were bought alive from different fishermen from different sites along the lake in spring 2018. Fish were collected in large aerated tanks filled with water from the lake and transported to the National Institute of Oceanography and Fisheries (NIOF), Physiology Laboratory. The fish rinsed with distilled water. Samples are dissected by a sterile knife on a clean glass surface and then divided into muscle and liver tissues. Within properly sterilized polythene bags, the split portions of each body are preserved. Bags were labeled and placed in the deep freezer at -25° C until various tests were carried out.

Estimation of trace elements

Fish tissue was separated and digested separately with a concentrated HNO3 and HClO4 by the ratio of 3:1. In a (100 ml) test tube, the specimen 0.5 g plus (10) ml of conc. HNO3 warmed at 100, 150, 200 and 250 ° C on a hot dish for one and a half hours. Then added (HClO4) to it. We then poured (2) mL of (1 N nitric acid) and put it on a hot dish and heated the mixture to fully digest and be transparent. These digested samples were transported to volumetric flasks (50 mL) plus the volume was made up by adding deionized water. Specimens filtered via a membrane filter (Type HV) of (0.45 µm) Millipore. This filtrate was examined for trace elements (Al, Cd, Pb, and, As) following the method of Andaleeb et al. (2008) by using Atomic Absorption Spectrophotometer Analyst 400-Perkin Elmer (USA). Hg was measured following the method of Larry et al. (1993). Digested 0.5 g of fish tissue then added 5 ml stannous chloride solution to reduce mercury to elemental form and then measured using Flameless Atomic Absorption Spectrophotometer equipped with mercury hydride system MHS -Cold Vapour Technique. Each sample was run in six replicates and the results were expressed in terms of µg/g wet weight (mean ± SE).

In determining trace element concentrations in fish samples, both samples and blanks were processed in the same way using the same reagents to minimize the error.

The chemical reagents and acids were an analytical grade (E. Merck, Germany) and the double deionized water also used for the preparation of all solutions. The standard solutions for calibrations of each element were prepared by diluting stock solutions of 1000 mg/l. The glass objects were soaked and rinsed for 24h, together with a 0.5% KMNO4 solution (w/v) and 10% of the nitric acid and washed into distilled water.

Estimation of risk assessment

The estimated weekly intake (EWI) of trace element through human consumption was compared with the current provisional tolerable weekly intakes (PTWI) for Hg, Al, Cd, Pb, and As (FAO/WHO 2004).

If the estimated value of EWI was lower than PTWI, this means that food consumption does not pose a significant health risk to the exposed population

According to General Authority for Fishery Resources Development, Ministry of Agriculture and Land Reclamation, Egypt, GAFRD (2017), and Central Agency for Public Mobilization and Statistics (CAMPAS 2017), the annual share per capita/Kg was 22.72 Kg/ year.

Estimated daily intake (EDI) = C × mean fish intake (g / day/capita).

Where C is the concentration of trace element in fish sample (μg/g wet weight)

Fish consumption / day/capita= 22.72×1000 ÷ 365= 62.25 g /day

Estimated weekly intake (EWI) = EDI × 7 days / week

EWI per body weight (kg) = EWI ÷ reference consumer body weight

(Assuming the body weight of an adult is 70 kg, the young person is 40 kg and the child is15 kg (Salas et al., 1985; Albering et al., 1999).

The percentage of trace elements intake to PTWI is calculated according to

%PTWI = (EWI÷PTWI) × 100

Where, %PTWI is the percentage of provisional tolerable weekly intakes can be calculated for each trace elements by the well-known weekly safety reference dose (PTWI) set by the joint (FAO/WHO 2004)

Maximum daily intake MDI (in grams) = is the estimated safety weekly intake of fishes (in grams) that children, young people and adult should consume less than it to attain the PTWI according to FAO/WHO (2004).

MDI =PTWI × BW ÷ C × 7

Where MDI is maximum daily intake; C is the element concentration (μg/g)

MWI= PTWI×BW / C

Where MWI is maximum weekly intake; C is the element concentration (μg/g)

Estimation of proximate body composition

In a glass homogenizer in 5ml saline, a specimen of 0.1 g of muscle was homogenized for 3 minutes, and then centrifuged for 10 minutes at 3000 rpm. The supernatant used to assess total protein content (Lowry et al. 1951); total lipid content calculated by Henry et al. (1974), and the method of Kemp et al. (1954) was used to assess total carbohydrates in tissues. The moisture content after drying at 100 °C ± 2 °C was considered as losses in sample mass (1 g). To assess the ash content, the residual mass was heated to 600 ° C ± 10 ° C (AOAC 2002).

Estimation of activity of antioxidant enzyme

The SOD (EC 1.15.1.1) activity was calculated by the enzyme's ability to inhibit the phenazine methosulphate–mediated reduction of nitroblue tetrazolium dye at 550 nm for 5 minutes (Nishikimi et al. 1972). The SOD activity is expressed as (U/g). GSH activity (EC 1.8.1.7) was documented using the methods of Beutler et al. (1963). The reduced chromogene can be estimated at 405 nm in direct proportion to the concentration of GSH, expressed as (mg / g). GPx (EC 1.11.1.9) activity measured using a substrate H_2O_2 via the method of Paglia and Valentine (1967), the reaction monitored indirect way at 240 nm as the rate of oxidation of NADPH to NADP+ for 3 minutes. Enzyme activity was expressed as (mU/mL). CAT activity (EC 1.11.1.6) was measured using the method of Abei (1984) by hydrolysis of H_2O_2 and the resulting decrease in absorbance at 510 nm. The results were expressed as (U/g). The GR (EC 1.6.4.2) activity was analyzed using the method of Goldberg and Spooner (1983) described the ability of the enzyme catalyzes the reduction process of GSSG to GSH when NADPH is oxidized to $NADP^+$ and calculated as (U/L) at 340 nm at 37°C

Statistical analyses

Using the Statistical Processor System Support (SPSS 20, Armonk USA), data are analyzed by the one way ANOVA, presented as mean ± standard error, values considered statistically at ($P < 0.05$) significant. Different superscripts are statistically significant ($p < 0.05$).

Results And Discussion

Trace element concentrations

The average concentrations of Hg, Al, Cd, Pb and As in *T. zillii and C. gariepinus* muscles collected from lake Edku are presented in Fig. (2) by mean ± SE expressed as (µg/g ww). The elements reported the descending order Hg< Al < Cd < Pb < As in *T.zillii* and *C. gariepinus*. The highest concentration was for As in *C. gariepinus* (1.54 ± 0.08 µg/g), but the lowest concentration was for Hg in *T.zillii* (0.47 ± 0.08 µg/g).

This study recorded an average Hg concentration in *C. gariepinus* (0.59 ± 0.07 µg/g) that was insignificantly greater than *T.zillii* (0.47±0.08 µg/g). The *f*-ratio's value was 1.20085; the *p-value* was 0.298842. In our results Hg in *T.zillii* and *C. gariepinus* exceeded the permissible limit EOSQC (1993) (0.50 ppm wt), FAO/WHO (1992) (0.50 ppm wt), EC (2006; 2008 amended 2011) 0.50 µg/g. Also, the results exceed the concentration of Hg (0.48 ppm) in *Oreochromis niloticus* from ten Egyptian governorates was recorded by Abdel-Mohsien and Mahmoud (2015). Abdel-Kader and Mourad, (2019a) demonstrated that Hg at *C. gariepinus* from Maryout lake ranged from 0.54 to 2.50 µg/g were much higher than our results. Whereas, the level of Hg was recorded in *T. zillii* 0.60±0.01 and *C. gariepinus* 0.73±0.07 µg/g from Burullus Lake, Egypt, was lower than our results (Abdel-Kader and Mourad, 2020). Symptoms of organic Hg toxicity include depression, memory problems, tremors, fatigue, hair loss and headache (Martin and Griswold 2009).

In this study, the average Al concentrations in the muscles of *C. gariepinus* was 1.05 ± 0.08 µg/g significantly ($p < 0.05$) higher than *T.zillii* 0.59 ± 0.12 µg/g (The *f*-ratio value is 9.4512. The *p*-value is 0.011755). *C. gariepinus* and *T.zillii* from Burullus Lake – Egypt with average 2.07± 0.1 and 1.85±0.1 µg/g, respectively were much higher than our results (Abdel-Kader and Mourad, 2020). Al in freshwater sources involves domestic waste, metal-related production processes, and discarded sewage sludge in freshwater sources (James 1991) caused physiological disorders.

This study showed that the mean concentration of Cd in C. *gariepinus* 1.13±0.08 μg/g was insignificantly higher than *T.zillii* 0.93 ± 0.05, (the *f*-ratio value = 3.74752 and the *p*-value = 0 .08164). So, the concentrations of Cd were reported in our results were largely above the permissible level of EOSQC (1993) (0.1 ppm), WHO (2006) (0.5 mg/kg), FAO (2010) (0.1 mg/kg), and EC (2006; 2008 amended 2011) 0.050 μg/g. The high load of Cd is due to different industrial and domestic channels induced in the Lake. Our results were greater than Saeed and Shaker (2008) who recorded that Cd (0.014 μg/g) in *O. niloticus* from Lake Burullus. Olabanji and Oluyemi (2014) recorded that Cd concentrations were 0.60 to 0.70 μg/g in *T. zillii* obtained from Opa Reservoir at Obafemi Awolowo University (OAU), Ile-Ife, Nigeria. Noor and Zutshi (2016) reported Cd muscle tissue accumulation 0.41 ± 0.50 μg/g of freshwater fish *Labeo rohita* from Bangalore, Karnataka. Cd at Maryout Lake ranged from 0.7 to 1.90 μg/g which much greater than our results (Abdel-Kader and Mourad 2019a). Abdel-Kader and Mourad (2020) recorded that Cd in *C. gariepinus* (1.66 μg/g) was insignificantly higher than *T. zillii* from Burullus Lake – Egypt. Cadmium (Cd) can cause anemia, osteoporosis, emphysema, non-hypertrophic, renal injury, and chronic rhinitis (Flora et al. 2008).

In this research, the concentration of Pb was insignificantly higher in *C. gariepinus* 1.31±0.08 with (*f*-ratio value = 2.19383, the *p*-value = 0.169371) than *T.zillii* 0.94 ± 0.08 μg/g. So , the Pb level in the muscles exceeded the permissible limit recommended by EOSQC (1993) (0.1 ppm w wt), FAO/WHO (1999) (0.214 μg/g wet weight) EC (2006; 2008 amended 2011) 0.30 μg/g. This study observed that Pb concentrations were much higher than those reported by Saeed and Shaker (2008) that reported Pb concentrations in *O. niloticus* collected from Lake Borollus was 0.016 μg/g. Olabanji and Oluyefmi (2014) reported that Pb levels were 1.80 ± 2.20 to 2.00 ± 0.17 mg/ kg at *T. zillii* from Opa Reservoir at Obafemi Awolowo University (OAU), Ile-Ife, Abdel-Kader and Mourad (2019a) showed that Pb concentration of *C. gariepinus* from Maryout Lake-Egypt 0.2 to 0.8 μg/g was lower than our results. Abdel-Kader and Mourad (2020) reported that Pb levels in *C. gariepinus* and *T. zillii* were 2.29 and 1.20±0.09 μg/g, respectively, from Burullus Lake – Egypt exceeded our the levels of our results**.** Tayel et al. (2007) suggested that high levels of lead may be due to motorboat traffic, Industrial discharges, mine and smelting operations. After exposure to Pb, a significant increase in immunological metrics, indicating that Pb may decrease the respiratory system, leading to increase susceptibility to disease (Flora et al. 2008).

The average concentration of As was insignificantly higher in *C. gariepinus* 1.54 ±0.14 μg/g with (*f-ratio* value = 0.21817, the *p-value* =0.650449) at $p < 0.05$ than *T.zillii* 1.43 ± 0.17 μg/g. The concentrations of As in tissues of two studied species were in the range of the permissible limit recommended by FAO/WHO (2004) (2.0 ppm) w wt. Abdel-Kader and Mourad (2019a) showed that As concentration of *C. gariepinus* from Maryout Lake-Egypt, 0.40 to 1.00 μg/g lower than our results. As levels in *C. gariepinus* and *T. zillii* from Burullus Lake – Egypt with average values of 2.03 ± 0.1 and 1.703 ± 0.1 μg/g, respectively, were much higher than our results (Abdel-Kader and Mourad 2020). Chronic exposures to trace elements result in the accumulation of the toxic elements lead to disease conditions (Authman et al. 2015).

Human risk estimation

Trace element levels in consumed fish can cause health hazards to humans (Castro-gonzález and Méndez-armenta, 2008). It is, therefore, essential to determine the concentration of elements in fish to indicate that it does

 the elements concentration constant below acceptable levels

(Palaniappan and Karthikeyan, 2009). As shown by PTWI for Cd, Pb, Hg, As and Al were 0.007, 0.025, 0.005, 0.015, and 1mg/kg, respectively (FAO/WHO, 2004).

Table 1 showed the calculated values for the estimated daily intake (EDI) (µg/kg body weight/day), estimated weekly intake (EWI) (µg/kg body weight/week), PTWI%, maximum daily intake (MDI), and maximum weekly intake (MWI) of Cd, Pb, Hg, Al, and As in the muscles of T.zillii and C. gariepinus from Edku Lake what should consume by children, youth, and adult..

This study showed that the EDI, EWI and PTWI % of elements by adults were lower than that was reported by the PTWI values established by FAO/WHO except for Cd residue in *C. gariepinus* recorded 100.42 PTWI %. So, the two fish species did not present a risk to adult health with advice on safe levels of consumption of *C. gariepinus* for adults. Table 1 showed that the EDI, EWI and PTWI% of two fish species muscles by the children and young people pose a health risk, especially intake of Hg, Cd and As in young people, plus Pb for children. In China, Yi et al. (2017) confirmed that the daily fish consumption recorded in the upper Yangtze River, has shown no adverse health effects on humans. In Maryout Lake—Egypt, Abdel–Kader and Mourad (2019a) showed that EWI of trace elements (Cd, Hg, Pb, and As) in *C. gariepinus* was safe for adults regards to the PTWI. Moreover, in Burullus Lake—Egypt, Abdel-Kader and Mourad (2020) demonstrated that EWI of trace elements (Cd, Hg, Pb, As, and Al) in fish species tilapia and catfish were accepted for adults and youth, unsafe for children.

In our results, Table 1 showed that the EDI and EWI of trace elements As> Pb >Cd > Al > Hg followed the ranking order of children >youth > adults based on consumption of *T.zillii* and *C. gariepinus* muscles. Besides, Table 1 showed that the PTWI% followed a ranking order of Cd >As> Hg>Pb >Al based on consumption of *T.zillii* whereas Cd >Hg >As> Pb >Al based on consumption of *C. gariepinus.* So, our study calculated the maximum safety grams of muscles for two fish species in a diet/ day or week. Thus, it recommended that the children should consume less than 16.12857 g/day or 112.90 g/week *T.zillii* muscle, and 13.27 g/day or 92.92 g/week *C. gariepinus* muscle. Whereas, Youth should consume less than 43.01 g/day or 301.7g/ week T.zillii and 35.39714 g/day or 247.78 g/week *C. gariepinus* muscle. Finally, adults should consume less than 23.1 g/day or 161.7 g/week *T.zillii* and 61.94286 g/day or 433.6 g/week *C. gariepinus* muscle.

Table 1 Estimated daily intake (EDI) (µg /day/capita), estimated weekly intake (EWI) (µg/week /c TWI%, maximum daily intake (MDI), maximum weekly intake (MWI) of Hg , Al ,Cd, Pb and As in muscl *illii* and *C. gariepinus* muscles consumed by a child 40 kg, young 40 kg, and an adult70 kg from Edku gypt.

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Fish species	Intake	Trace elements				
		Hg	Al	Cd	Pb	As
T. zillii consumed by a child 15 kg)	*EDI*	1.9505	2.44	3.858	3.9	5.93
	EWI	13.65*	17.08	27.01*	27.3*	41.51*
	PTWI	5.0	1000	7.0	25.0	15.0
	PTWI%	273	1.708	385.8571	109.2	276.7333
	MDI	22.79571	3631.957	16.12857	56.99	22.79571
	MWI	159.57	25423.7	112.90	398.93	157.3
ariepinus consumed by child15 kg)	*EDI*	2.44	4.357143	4.688571	5.545714	6.39
	EWI	17.08	30.50	32.82*	38.82*	44.73*
	PTWI	5.0	1000	7.0	25.0	15.0
	PTWI%	341.6	3.05	468.8571	155.28	298.2
	MDI	18.15857	2040.814	13.27429	40.89286	20.87143
	MWI	127.11	14285.7	92.92	286.25	146.10
T. zillii consumed by (young 40 kg)	*EDI*	0.73	0.917143	1.447143	1.461429	2.224286
	EWI	5.11*	6.42	10.13*	10.23	15.57*
	PTWI	5.0	1000	7.0	25.0	15.0
	PTWI%	102.2	0.642	144.7143	40.92	103.8
	MDI	60.79	9685.229	43.01	151.9714	60.79
	MWI	425.53	67796.6	301.07	1063.8	419.5
C. epinus consumed by (young40 kg)	*EDI*	0.917143	1.632857	1.757143	2.037143	2.395714
	EWI	6.42	11.43	12.30*	14.26	16.77*
	PTWI	5.0	1000	7.0	25.0	15.0
	PTWI%	128.4	1.143	175.7143	57.04	111.8
	MDI	48.42571	5442.171	35.39714	109.05	55.65857
	MWI	338.98	38095.2	247.78	763.35	389.61
T. zillii consumed by *an adult 70kg)*	*EDI*	0.41	0.524286	0.825714	0.835714	1.271429
	EWI	2.87	3.67	5.78	5.85	8.90
	PTWI	5.0	1000	7.0	25.0	15.0
	PTWI%	57.4	0.367	82.57143	23.4	59.33333
	MDI	106.3829	16949.14	75.26857	23.1	104.8943
	MWI	744.68	118644.0	526.88	161.7	734.26
C. epinus consumed by an adult 70kg)	*EDI*	0.524286	0.932857	1.004286	1.164286	1.368571
	EWI	3.67	6.53	7.03*	8.15	9.58
	PTWI	5.0	1000	7.0	25.0	15.0
	PTWI%	73.4	0.653	100.4286	32.6	63.86667
	MDI	84.74571	9523.714	61.94286	190.7143	97.40143
	MWI	593.22	66666	433.6	1335	681.81

Proximate body composition

Fish muscle included moisture, proteins, carbohydrates, and fats considers as main components (Love 1980). In this analysis, mean values ± SE are shown proximate body composition in (Fig. 3) at *T. zillii* and C*. gariepinus* muscle tissue. This analysis showed a significant difference ($P < 0.05$) in the comparison of mean carbohydrate values (mg/g) for two fish species.

In this study, *T. zillii* recorded the maximum mean carbohydrate value 16.11± 0.67 while *C. gariepinus* recorded the lowest amount of carbohydrate 11.28± 0.68. In contrast, carbohydrates of *T. zillii* 14.1±0.7 mg/g, was higher

 Lake (Abdel-Kader and Mourad 2020). Carbohydrate values in tilapia are insignificant higher than crab values. In the lobster, carbohydrate is significantly lower than the other two species (Ayanda et al. 2018).

In this research, lipid content recorded an insignificant difference in two fish species. There was an increase in lipid content (mg/g) observed in the *T. zillii* (24.21±0.93) followed by *C. gariepinus* (21.96±0.95 mg/g) for the minimum value. Sobha et al. (2007) observed a reduction in lipid content following the exposure of *Catla catla* to Cd; *Perca flavescens* (Levesque et al. 2002) and *Anguilla anguilla* (Pierron et al. 2007). The lipid content may be affected by different factors such as feed structure, geographic origin, age difference, reproductive stage, and catching season (Murillo et al. 2014).

A comparison of the means of protein (mg/g) showed a significant difference ($P < 0.05$) for two fish species. *C. gariepinus* showed the maximum mean protein value 64.33±1.01 followed by *T. zillii* 55.66±1.95. It may also be caused by heat shock protein production or toxic free radicals, or apoptosis may be caused by elements. Selvam et al. (2014) reported a significant difference ($P<0.05$) in the protein content in Swat Khyber Pakhtunkhwa Pakistan between five different commercial *Cyprinida* fish species. Palaniappan and Vijayasundaram (2009) noted a significant decrease in protein and lipid content in *Labeo rohita* upon As exposure. Depletion of the lipid and protein content of Pb-exposed *Catla catla* muscles (Palaniappan et al. 2008), can be caused by tissue organization and its use in lipoprotein cell repair, essential cellular elements of cytoplasmic cell membranes and organelles (Filipovic and Raspor 2003). Abdel-Kader and Mourad (2019b) showed an increase in protein content from 56 to 27 mg/g and 26 to 32 mg/g in lipid content from two basins at Maryout Lake. Abdel-Kader and Mourad (2020) showed that the protein 57.16± 2.6 mg/g and lipid content 11.98±1.1 mg/g of *T. Zillii* were greater than of *C. gariepinus* 56.16±3.0 mg/g and 11.65±0.7 mg/g, respectively, from Burullus Lake, Egypt

In this study, moisture content's mean recorded an insignificant difference ($P >0.05$) for the two fish species. *T. zillii* showed the highest mean moisture content 78.33±1.58% while the least mean moisture content was recorded in the *C. gariepinus* 77±0.45%. Water content in *C. gariepinus* ranged from 75±4.40 to 53.00 ±7.70 % from two basins at Maryout Lake (Abdel-Kader and Mourad 2019b) which lower than moisture content of this study. Abdel–Kader and Mourad (2020) showed that the water content in *T. zillii* 64± 1.9% and *C. gariepinus* 64 ± 2.1% from Burullus Lake, Egypt much lower than water content in our study. Ayanda et al. (2018) recorded a significant rise in water content in tilapia from Nigeria at Makoko River may be rendering the fish sensitive to microbial spoilage, damage to fatty acids, and consequently decreasing fish quality thereby reducing its survival time (Omolara and Omotayo 2009)

In our results, fish species showed an insignificant decrease in the ash content. Maximum ash content was observed in the *T. zillii* (7.33±1.00%) while the minimum value (6.66±0.85 %) was recorded in the *C. gariepinus*. In contrast, the ash content was significantly reduced in tilapia from Nigeria at Makoko River (Ayanda et al. 2018). Finally, Abdel-Kader and Mourad (2020) recorded that the ash content of *T. zillii* 14.25±1.7% and 3.37± 0.1% of *C. gariepinus* from Burullus Lake, Egypt.

Antioxidant enzyme activity

Trace elements such as Cd, Pb, Hg, As and Al can inhibit cellular antioxidant activity, particularly thiol-containing antioxidants and enzymes (Sevcikova et al. 2011). Trace elements can promote oxidative damage by directly increasing the cellular concentration of ROS and altering the cellular antioxidant capacity in fish (Sevcikova et al. 2011).

This study recorded a non-significant decrease in antioxidant SOD, GPx, and GR demonstrated in the liver, whereas, CAT and GSH were significant ($P < 0.05$) (Fig. 4) by mean values ± SEM. GSH, CAT, GPX, and GR activity in *C. gariepinus* 41.66 ± 2.01mg/g, 21.66 ± 2.6 U/g, 36.5 ± 3.01 mU/L and 41.16 ± 1.0 U/L, respectively, to be less than *T. zillii* 47.83 ± 3.6 mg/g, 38.16 ± 4.2 U/g, 45.33 ± 4.5 mU/l, and 41.83 ± 3.4 U/L, respectively. SOD activity in *T. zillii* 47±3.5 U/g was less than *C. gariepinus* 48.16 ± 4.23 U/g (Fig. 3).

After exposure to Pb, a significant reduction in CAT activity was observed in *O. niloticus* (Elarabany and Bahnasawy 2019). CAT activity reduced due to the generation of superoxide radicals that decrease CAT activity sharply (Ahmad 2000). When hydrogen peroxide changing to O_2 and H_2O, CAT protects fish from oxidative stress (Atli and Canli 2007). Decreased GR activity may deplete of GSH if additional GSH synthesis cannot occur to protect its redox status, while an increase in activity may be related to the re-establishment of oxidized GSH (Atli and Canli 2007). An insignificant difference in GR activity was found in *Mugil* sp (Rodriguez-Ariza et al. 1993) which was sampled from contaminated pesticide areas. Abdelazim et al. (2018) recorded the recognized decrease in GR following exposure of the Nile tilapia to zinc oxide nanoparticles. GSH is a potent antioxidant capable of binding freely to radicals and trace elements, such as Cd and Pb, and defending cells against their adverse effects (Jan et al. 2011). GSH induction is the primary step in the defense system to keep against oxidative stress; it showed that metal induces increased hepatic GSH synthesis (EL-Gazzar et al. 2014). Trace elements respond to GSH thiol groups that can cause depletion of GSH then oxidative tissue stress in the tissue (Stohs and Bagchi 1995). Monteiro et al. (2010) observed oxidative stress followed exposure to Hg rely on that the metal-binding GSH reduce toxicity (Elia et al. 2003). GSH levels in zebrafish decreased after dimethoate exposure (Ansari and Ansari 2014), in juveniles after Pb exposure (Saliu and Bawa-Allah 2012). The liver is the main target organ of GPx production for metabolized oxidants. That indicates the sensitivity to the oxidative stress to which the fish has been exposed (Lenartova et al. 1997). GPx detoxifies H_2O_2 or organic hydroperoxides produced in the LPO (Halliwell and Gutteridge 1999). Cadmium caused a significant reduction in a hepatic GPx activity in *niloticus* after 42 days of exposure suggested that GPx had a tissue-specific response to Cd exposure and depended on tissue-specific alternations of CAT and SOD documented in *O. niloticus* exposed to diazinon (Durmaz et al., 2006). Cd induced deactivated GPx activity with a great affinity to bind with –SH groups, and so, initializes the activity of the enzyme (Giguère et al. 2005). Orun et al. (2008) showed significant alterations of GPx activity in fish tissues *Onchorhynchus mykiss* after Cd exposure. The direct effects of the element on enzyme molecules at the active site could be due to a reduction in GPx, although an increase in GPx activity may be attributed to the fish’s stress. A substantial increase in SOD production in the liver, when exposed to a mixture of both elements may be attributable to the summation effect of both elements resulting in the accumulation of free radicals where SOD acts as an antioxidant enzyme protective (Asagba and Obi 2000). Barata et al. (2005) also observed diminished SOD and CAT activity after Cd exposure in *Daphnia magna* suggested that toxicants could produce several antioxidant/ prooxidant reactions because they are capable of ROS. Vinodhini and Narayanan (2009) recorded decreases in GPX, SOD and CAT after 32 days of exposure to Pb, Cd, Cr and Ni in *Cyprinus carpio* L. Abdel-Kader and Mourad (2019b) recorded decreases in the activity of CAT, GPX and SOD in *C. gariepinus* from the main basin at Lake Maryout. Abdel-Kader and Mourad (2020) recorded decreases in CAT, GR, GSH, GPX, and SOD activity of *C. gariepinus* followed by *T.zillii* .

Conclusion

In summary, high protein content was recorded in *C. gariepinus* followed by *T.zillii. Whereas* the high means of carbohydrates, fats, ash, and water content were recorded in *T.zillii* followed by *C. gariepinus*. SOD, GR, and GPx activity in the liver of *C. gariepinus* showed insignificant increases, whereas, the increase in CAT and GSH were significant. This study revealed that all toxic elements in *C. gariepinus* were consistently high, the highest element was As recorded in *C. gariepinus*, while, the lowest element was Hg in *T.zillii*. Although the trace element level concentrations exceeded the permissible levels suggested by Egypt, FAO, WHO, and EC in two fish species, the estimated weekly intake of Pb, Hg, As, and Al was below the PTWI except for Cd of *C. gariepinus* for adults, So, consumption of collected fish from Edku lake was safe for adults (70 kg) with advice on safe levels of consumption for *C. gariepinus* for adults , whereas unsafe for young people (40 Kg) and children (15 Kg) concerning the PTWI. The EDI and EWI of As> Pb >Cd > Al > Hg followed the ranking order of children >youth > adults based on consumption of *T.zillii* and *C. gariepinus* muscles. In addition, the PTWI% followed a ranking order of Cd >As> Hg>Pb>Al based on consumption of *T.zillii* whereas Cd >Hg >As> Pb >Al based on consumption of *C. gariepinus*. So, we recommended that the children should consume less than 16.12857 g/day or 112.90 g/week *T.zillii* muscle, and 13.27 g/day or 92.92 g/week *C. gariepinus* muscle. Youth should consume less than 43.01 g/day or 301.7g/ week *T.zillii* and 35.39714 g/day or 247.78 g/week C*. gariepinus* muscle. Finally, adults should consume less than 23.1 g/day or 161.7 g/week *T.zillii* and 61.94286 g/day or 433.6 g/week *C. gariepinus* muscle.

Also, the consumers should consider that the intake of two types of fish species in one meal increases the quantities of trace elements consumption. Accordingly, this study reply to the question in the title research, that, Egyptians will be at risk if they consume more than the maximum daily or weekly intake of Cd, Pb, Hg, As and Al through consumption of *T.zillii* and *C. gariepinus* in respective to the PTWI

Declarations

Author contributions

Mohamed H. Mourad was responsible for the general supervision of the research and contributed to the final review of the research before sending the manuscript to the journal for publication.

Heba H. Abdel-Kader dissected the fish and kept them in sterile bags, and marked and placed them in a deep freezer until analysis. She did the analyzes including the statistical analyzes, collected literature, wrote the manuscript, linguistically reviewed it, including the grammar, and sending the manuscript to the journal for publication.

Afaf Mohamed M. Haredi collected fish samples from the fishermen and assisting in dissecting and keeping them in sterile bags in a deep freezer.

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Data availability

The datasets used and/or analyzed during the current study are available from the corresponding author on

Compliance with ethical standards

Conflict of Interest

The authors declare that they have no conflict of interest.

Consent to participate and consent for publication

Not applicable

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Figures

Figure 1

Location map of the study area. Note: The designations employed and the presentation of the material on this map do not imply the expression of any opinion whatsoever on the part of Research Square concerning the legal status of any country, territory, city or area or of its authorities, or concerning the delimitation of its frontiers or boundaries. This map has been provided by the authors.

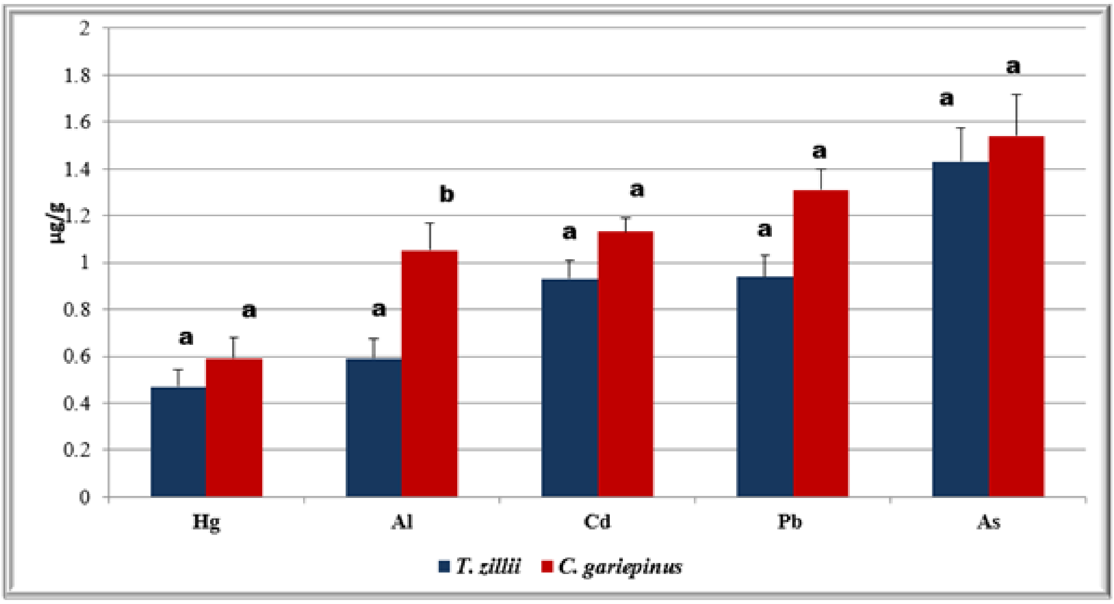

Figure 2

The average of trace element concentrations (µg/g wet weight) ± standard error in the muscles of T.zillii and C. gariepinus, (n=6). Different superscripts are statistically significant p < 0.05.

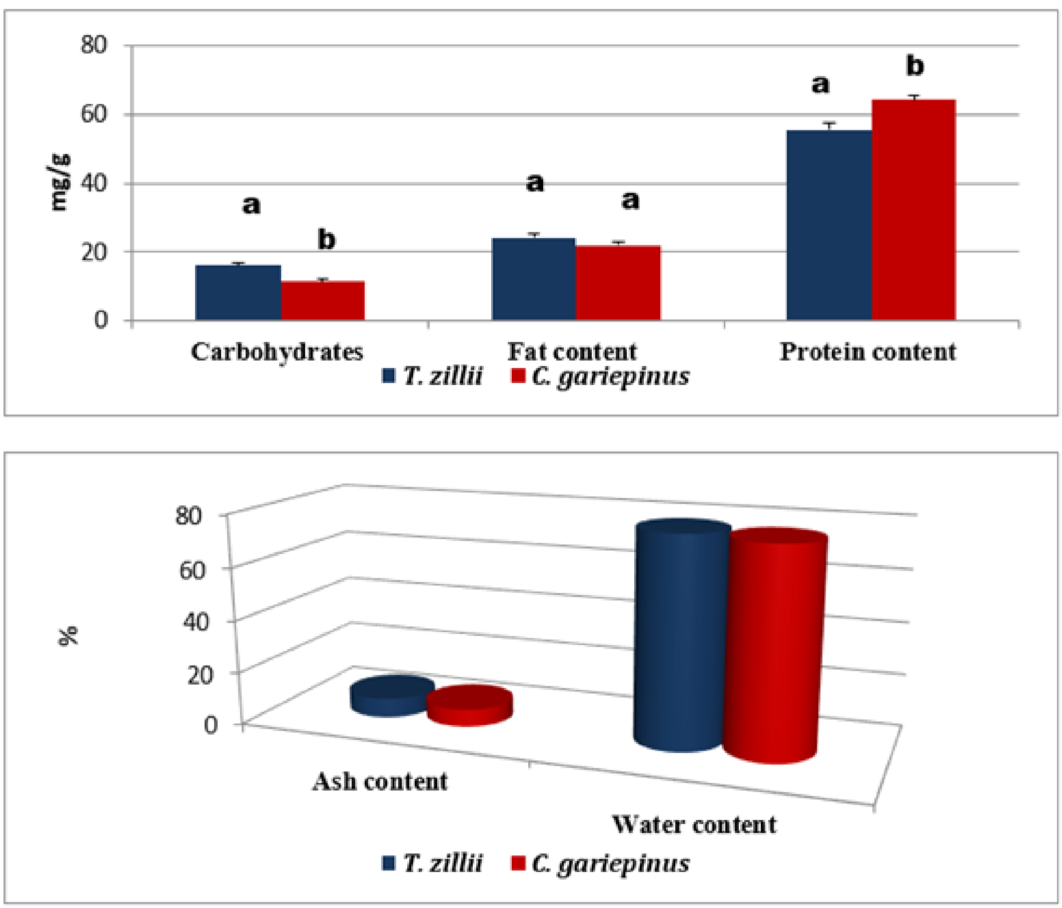

Figure 3

Proximate body composition in muscle tissue of T.zillii and C. gariepinus. (n = 6). Different superscripts are statistically significant $p < 0.05$

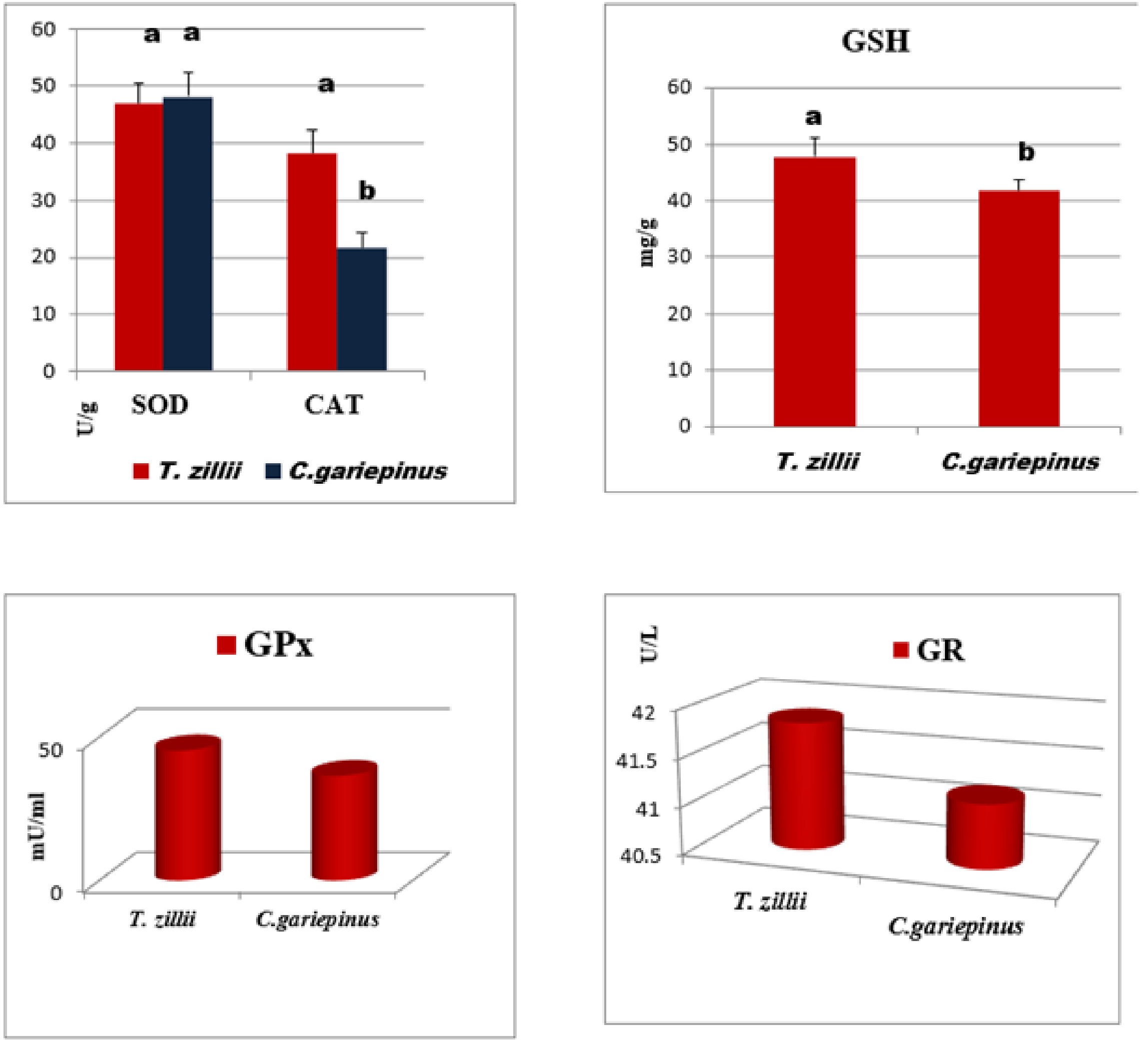

Figure 4

Antioxidant enzyme activity in the liver tissue of T. zillii species and C. gariepinus (n = 6). Different superscripts are statistically significant $p < 0.05$.